

# The potential efficiency of irrigation management and propargyl bromide in controlling three soil pests: *Tylenchulus semipenetrans*, *Fusarium oxysporum* and *Echinochloa crus-galli*

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**Abstract:** Propargyl bromide (3-bromopropyne, 3BP) is a potential alternative for methyl bromide. Little information is available about its efficiency in controlling pests. The purpose of this paper is to estimate the 3BP dose required for killing three pests and to compare the efficiency of water management approaches to that of fumigation. The pests, *Fusarium oxysporum* Schlecht (fungus), *Echinochloa crus-galli* (L) Beauv (grass) and *Tylenchulus semipenetrans* Cobb (nematode) were exposed to different 3BP concentrations in a sandy loam at 30 °C in a closed system. The lethal dose for killing 90% of the population (LD<sub>90</sub>) was calculated from the total applied mass, and varied from 0.3 µg g<sup>-1</sup> soil for the nematode, 3 µg g<sup>-1</sup> for the grass, and 9 µg g<sup>-1</sup> for the fungus. The concentration–time index for killing 90% of the population (CT<sub>90</sub>) was 11 µg g<sup>-1</sup> h for the nematode, 112 µg g<sup>-1</sup> h for the grass and 345 µg g<sup>-1</sup> h for the fungus. 3BP seems as efficient as other fumigant alternatives in controlling these pests. Using an open system, it was shown that the volume of soil in which the pests were controlled varied for different irrigation managements. Even 96 h after fumigation (with a concentration 10 times higher than would potentially be applied in the field), more than 20% of the soil volume had not reached the fungus and grass CT<sub>90</sub> of the non-irrigated soil. The soil underneath the furrow and the bed reached CT<sub>90</sub> only slowly in all irrigated treatments even though techniques for increasing efficiency were used (tarping, surface sealing with water and high application rate).

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**Keywords:** fumigant; nematodes; weeds; CT index; LD<sub>50</sub>; LD<sub>90</sub>; 3-bromopropyne; irrigation

## 1 INTRODUCTION

Methyl bromide is a class 1 stratospheric ozone-depletion chemical which will be phased out in 2005 as a result of the Montreal protocol. There is an urgent need to develop alternatives for methyl bromide, a highly efficient fumigant with broad-spectrum activity, high diffusion allowing penetration into non-accessible locations and short application time. Propargyl bromide (3-bromopropyne, 3BP) has interesting physical and chemical properties (Table 1), much like those of methyl bromide, for use as a soil fumigant.<sup>1</sup> 3BP has a simple structure similar to methyl bromide, but decays significantly more quickly, making it more environmentally friendly. However, little information is available on how pests respond to 3BP fumigation.

The pest response to a pesticide is described in terms of a dose that is lethal to a certain percentage of the pest population. This dose often refers to a concentration that kills 50% of the pest population (LD<sub>50</sub>) or 90% of the pest population (LD<sub>90</sub>). Ma *et al*<sup>2</sup> found that the concentrations of 3BP required for 50% inhibition of barnyard grass (*Echinochloa crus-galli* (L) Beauv) seed germination ranged from 2.8 to 48 g kg<sup>-1</sup> soil dry weight and from 11.2 to 182 g kg<sup>-1</sup> for control of *Fusarium oxysporum* Schlecht. The higher values corresponded to a soil containing 78% organic matter.

Pests react not only to the concentration but also to the duration of exposure. Goring<sup>3</sup> suggested calculating the actual dosage as the product of the concentration and time because a small dosage over a long period of time may control the pest as well as a

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**Table 1.** Propargyl bromide characteristics in Arlington sandy loam

Characteristic	Value	Reference
$\rho$ (g cm <sup>-3</sup> )	1.585	22
Molecular mass	119	22
Boiling point (°C)	89	30
Vapor pressure (MPa)	0.237	30
Solubility in water (mg liter <sup>-1</sup> )	14 800	22
$K_H^2$	0.05	22
$D_g^a$ (cm <sup>2</sup> h <sup>-1</sup> )	$1.76 \times 10^{-6}$	31, 32
$D_l^{water}$ (cm <sup>2</sup> h <sup>-1</sup> )	$1.76 \times 10^{-10}$	31, 32
Degradation rate, $k$ (h <sup>-1</sup> )	0.002–0.0075, 0.117	2, 17, 22
Half-life, $t_{1/2}^{soil}$ (h)	96–276	17, 22
Half-life, $t_{1/2}^{water}$ (h)	1128–1536	17, 22
$K_d^{soil}$ (cm <sup>3</sup> g <sup>-1</sup> )	0.084–0.96	22, 31

shorter exposure to a high concentration. He suggested that the concentration–time was constant for a pest regarding a specific pesticide and could be used as an index (CT) which can be calculated as follows:

$$CT = \int_0^t C_T(t) dt = B \quad (1)$$

where  $C_T(t)$  is the total concentration of all phases at time  $t$  (h) after fumigation.  $B$  is a constant. It may be argued that  $CT = B$  is not valid for most pest–pesticide combinations. Other models have been proposed<sup>4</sup> such as  $C^n T = B$  or<sup>5</sup>  $CT^a = B$ , while others<sup>6</sup> include a threshold in their model as  $(C - C_0)^n(T - T_0) = B$ . Abdalla and Lear<sup>7</sup> studied the control by methyl bromide of six nematode species at different concentrations, times and temperatures. They concluded that  $CT = B$  was valid under their experimental conditions. This latest model was therefore chosen because the experimental conditions of Abdalla and Lear<sup>7</sup> were the closest to the conditions used for this study.

This model can be used for different soil phases. Volatile pesticides partition in the soil into liquid, solid and gas phases. Pests may react to gas and/or liquid phases. The most effective phase in controlling a pest depends on the mode of action. It is likely that liquid phase exposure would be more damaging to an organism living entirely in the liquid phase. Many researchers<sup>2</sup> assume that nematodes respond only to the liquid phase exposure of fumigants because it is believed that their body must be surrounded by liquid so that their cuticle stays hydrated.<sup>3</sup> Fumigants in grain containers are often applied in very dry conditions. The fumigation seems to act in the gas phase by diffusing through the seeds. Therefore, the contribution of the different phases in controlling pests may vary from one pest to another, depending on the mode of action.

One would expect that the mode of action of 3BP would be similar to that of methyl bromide since the two fumigants have a similar structure. Methyl bromide has been widely used and studied,

but its mode of action is uncertain.<sup>8</sup> The molecule would alkylate DNA and proteins. DNA methylation inhibits replication and transcription. Methylation of SH groups such as SH-containing enzymes would likely be linked to the inhibition of dehydrogenase leading to ATP depletion and causing deleterious respiratory effects in the organism.<sup>8</sup> Thus the gas phase would be important for the air-breathing pests. 3BP does not have a methyl group. The mode of action must be different in some respects but it probably alkylates DNA. Early studies on methyl bromide have suggested that the release of inorganic bromide, an ion also released by 3BP, might be toxic. If this ion is partly responsible for the action, then the liquid phase would be important. However, there is no clear evidence that inorganic bromide causes deleterious effects on pests. Although it is unclear whether pest control by methyl bromide is predominately due to liquid or gas phase exposure, some studies<sup>5,7,9–13</sup> have calculated fumigant exposure on the basis of the gas phase concentration of methyl bromide. Other researchers<sup>14</sup> considered the effective dose as the summation of both the liquid and gas phases. For comparison purposes, and because the effective phase of 3BP is unknown, LD<sub>50</sub>, LD<sub>90</sub> and CT<sub>90</sub> indexes are discussed in this paper using the total applied concentration as well as the concentrations in the gas and the liquid phases, separately.

If 3BP were to be used as a soil fumigant, better management practices, in addition to lethal dose, would be needed to increase efficiency and decrease environmental impacts. A few techniques have already been suggested for other fumigants: (1) Increased moisture at the soil surface minimizes fumigant losses to the atmosphere.<sup>15</sup> (2) Fumigation in a wet soil may be more efficient than in dry soil in several conditions and for several species.<sup>3</sup> (3) Surface tarping with plastic reduces volatilisation and improves fumigation efficiency by increasing exposure time in the profile.<sup>16</sup> (4) Increased soil moisture accelerates 3BP degradation rate.<sup>17</sup> However, to save money, growers have a tendency to cover only the bed with plastic instead of covering the entire soil surface. (5) It has been shown that an initially very dry soil partially covered with plastic resulted in a very low 3BP concentration in the profile.<sup>18</sup> Many irrigation-tarping managements have not yet been tested, especially for 3BP. A combination of partial surface tarping with furrow irrigation, such as is done in the present work, is a potential option for increasing exposure time, accelerating degradation and decreasing losses to the atmosphere.

The objectives of this study were to (1) estimate LD<sub>50</sub>, LD<sub>90</sub>, and CT indexes for *F. oxysporum* (fungus), *E. crus-galli* (grass) and *Tylenchulus semipenetrans* Cobb (nematode) in a sandy loam soil and (2) compare the effect of different irrigation managements on the extent of the pest control zone in the soil profile.

## 2 MATERIAL AND METHODS

Two independent studies were conducted. The first one corresponded to the first objective. It was completed with the organisms in a closed system. The second experiment corresponded to the second objective. It was conducted in an open system with a large soil column.

### 2.1 Soil and fumigant

Arlington sandy loam, coarse loamy mixed, thermic, Haplic, Durixeralf, was sampled at the University of California Experimental Station in Riverside. The 2-mm sieve soil was sterilized and brought to a volumetric water content of  $0.17 \text{ cm}^3 \text{ cm}^{-3}$  which corresponded approximately to a 60% field capacity. This water content corresponds to the threshold for triggering the start of irrigation in different productions. Four 3BP (Albermarle Corporation) formulations were compared. Each formulation contained 74.9% of 3BP and the remaining 25.1% was composed of long-chain hydrocarbons, natural oils and other additives. The additives acted as stabilizers in the fumigant. The differences in pest response to the formulations were limited, except for the nematode. For this last species, there was 15% difference between the  $\text{LD}_{50}$  values of the least and most active formulations. This difference is insignificant compared with the differences observed between pests and between field studies in general. Therefore, only the values corresponding to the most efficient formulation are discussed. All values are corrected for the active ingredient content.

### 2.2 Pest lethal dosage

*Tylenchulus semipenetrans* were extracted from citrus roots. *Fusarium oxysporum* was isolated from *Heterodera schachtii* Schmidt nematode cysts. *Fusarium oxysporum* was chosen for its pathogenicity to a large range of hosts and its apparent high resistance to fumigation.<sup>5</sup> Both pests were obtained from the field described above. *Echinochloa crus-galli* seeds were purchased from Valley Seed Service, Fresno, CA. A germination test at 22 °C demonstrated that more than 99% of the untreated seeds were viable.

The nematodes were mixed with 50 g of soil (on a dry weight basis) in 165-ml serum bottles and incubated at 30 °C. After 16 h of incubation, each bottle was spiked with one of eight concentrations: 0.0, 0.1, 0.23, 0.36, 0.5, 0.8, 1.0 and  $3.0 \mu\text{g g}^{-1}$  soil ( $23.4 \text{ kg ha}^{-1}$  assuming soil density of  $1300 \text{ kg m}^{-3}$  and a depth of 0.6 m). The bottles were immediately capped and sealed with a Teflon-faced butyl rubber septum. The bottles were incubated at  $30 (\pm 0.5) ^\circ\text{C}$  for 48 h. This temperature was chosen to represent typical soil temperature in the upper crop zone during fumigation in California and Florida. Additional bottles containing nematodes, but not fumigated, were used as control treatments. The Baermann funnel method<sup>12</sup> was used to extract the nematodes from the soil. In short, the soil was transferred into a 3-mm mesh screen on which a paper filter had been placed

to retain the soil while allowing the nematodes to pass through it. The collected motile nematodes were counted using a dissecting microscope.

The materials and methods for fungi and grass were adapted from Ma *et al.*<sup>2</sup> Fungi were grown on potato dextrose agar (PDA) and transferred into 500-ml Erlenmeyer flasks containing 500 g of millet seeds and incubated for 2 weeks at 30 °C. Millet seeds were chosen because they seem resistant to fumigation while being good hosts for the fungi. Millet seeds were also used for the same fungus by Hutchinson *et al.*<sup>19</sup> and for other fungi.<sup>14</sup> To obtain uniform inoculation, the seeds were shaken every 2–3 days. The millet seeds were then dried in a laminar-flow hood under sterile conditions for 24 h and stored in clean plastic bags at 5 °C before use. Thirty millet seeds were mixed with 50 g of soil. The soil was then spiked with one of nine concentrations: 0.0, 4.0, 6.0, 8.0, 9.0, 10.0, 11.0, 14.0 and  $20.0 \mu\text{g g}^{-1}$  soil ( $156 \text{ kg ha}^{-1}$  using the same assumptions as above). The jars/bottles were immediately capped and sealed with Teflon-faced butyl rubber septa and incubated at  $30 (\pm 0.5) ^\circ\text{C}$  for 48 h. The seeds were recovered from the soil by sieving. Thirty seeds were randomly selected and transferred to PDA medium in a sterile Petri dish at 30 °C. Fungus growth was monitored daily. The colonies (infected seeds) were counted after 5 days. Fumigated seeds that were not infected showed more than 99% viability.

The efficiency of fumigation was expressed as the percentage germination inhibition compared to the non-fumigated treatment, as used by other researchers.<sup>9,20</sup> The number of individual chlamydospores formed on seed surface was not used as an inoculum unit because the purpose of fumigation is to reduce pathogen pressure sufficiently so that plant can grow without adverse impact. The complex seed–fungus is an indicator of the fumigation efficiency. It integrates the factors influencing sterilization/fumigation. In addition, Minuto *et al.*<sup>9</sup> showed that a minimal number of colony-forming units needs to be present to cause plant disorder, and decided to use the percentage of emerged plants as an indication of the control of *Fusarium* by fumigation. Thus, the lethal dosage ( $\text{LD}_{50}$ ,  $\text{LD}_{90}$  and  $\text{CT}_{90}$ ) presented for the fungus does not exactly correspond to the fungus population killed but instead to the efficiency of fumigation in reducing the negative impact of fungi on plant growth.

Fifty barnyard seeds were mixed with 50 g of soil and incubated at 30 °C for 48 h in the same type of container. Fumigation consisted of an application of one of nine concentrations: 0.0, 1.0, 2.0, 2.5, 3.0, 3.5, 4.0, 6.0 and  $8.0 \mu\text{g g}^{-1}$  soil ( $62.4 \text{ kg ha}^{-1}$  using the above assumptions). The containers were immediately sealed and incubated at  $30 (\pm 0.5) ^\circ\text{C}$  for 48 h. Thirty barnyard seeds were randomly removed from the soil by sieving, transferred to a Petri dish containing a moist germination blotter, and, after 5 days incubation at 30 °C, the germinated seeds were counted.

The general form of pest survival as a function of the applied concentration was calculated as follows:<sup>21</sup>

$$S = C_0 + \frac{(a - C_0)}{1 + (C_T/C_{50})^b} \quad (2)$$

$S$  is the pest survival (%).  $C_T$  is the total 3BP concentration ( $\mu\text{g g}^{-1}$ ).  $C_{50}$  is the soil concentration ( $\mu\text{g g}^{-1}$ ) corresponding to nearly  $\text{LD}_{50}$ , and  $C_0$  denotes the minimal concentration threshold ( $\mu\text{g g}^{-1}$ ).  $C_0$ ,  $C_{50}$ ,  $a$ , and  $b$  are fitted parameters. For  $\text{LD}_{50}$ ,  $S = 50$  and for  $\text{LD}_{90}$ ,  $S = 10$ . The CT index was calculated as in eqn (1).

In this experiment, the concentration of 3BP over time was not monitored. This is usually done in this laboratory and the degradation rate in this soil under these conditions is known. Propargyl bromide degrades in Arlington Sandy loam at a rate,  $k$ , varying<sup>2,22</sup> from  $0.0075 \text{ h}^{-1}$  to  $0.0117 \text{ h}^{-1}$  depending mostly on temperature and soil desinfection. For this experiment, a  $k$  value of  $0.0117 \text{ h}^{-1}$  was used because the conditions were the same as in Ma *et al.*<sup>2</sup> Water content and temperature remained constant throughout the experiment. Since exposure time (48 h) is known,  $C_T(t)$  can be estimated using the first-order rate constant ( $k$ ) as follows:

$$-C_T(t) = C_i e^{-kt} \quad (3)$$

where  $C_i$  is the initial concentration ( $\mu\text{g g}^{-1}$ ). Integrating eqns (3) into (2) and solving, we get:

$$C_T(t) = C_i(1 - e^{-kt})/k \quad (4)$$

Applied concentrations in liquid  $C_l$  ( $\mu\text{g cm}^{-3}$ ) and in gas  $C_g$  ( $\mu\text{g cm}^{-3}$ ) phases were calculated using Henry's law constant<sup>22</sup>  $K_H \approx 0.05$ , ie assuming equilibrium between the phases (Table 1). The relationships between the total concentration,  $C_T(t)$  ( $\mu\text{g cm}^{-3}$ ), and the other phases are:

$$C_l = C_T(t)/(K_d \rho_a + \theta_w + K_H \theta_a) \quad (5)$$

$$C_g = K_H C_l \quad (6)$$

$$C_s = K_d C_l \quad (7)$$

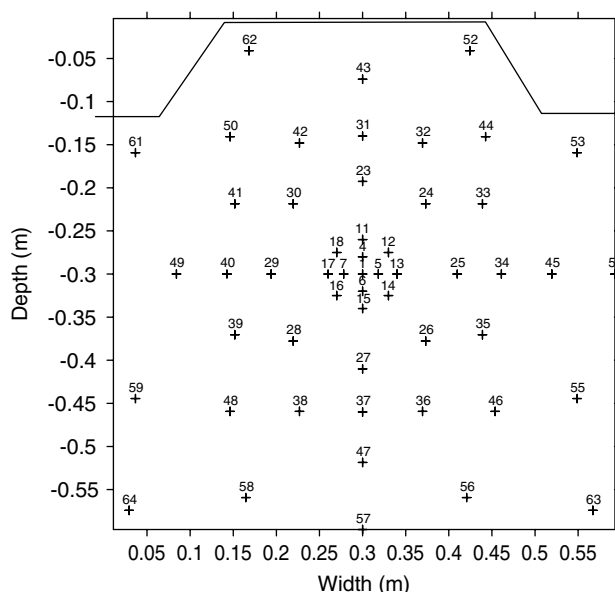
where  $K_d$  is the sorption coefficient<sup>2</sup> ( $0.96 \text{ cm}^3 \text{ g}^{-1}$ ),  $\theta_w$  is the soil volumetric water content ( $0.17 \text{ cm}^3 \text{ cm}^{-3}$ ) and  $\theta_a$  is the air content ( $0.84 \text{ cm}^3 \text{ cm}^{-3}$ ). This high air content  $\theta_a$  includes the air in the soil and the air in the vials, since the soil did not occupy the entire vial volume.  $C_s$  is the sorbed concentration ( $\mu\text{g g}^{-1}$ ) and  $\rho_a$  is soil bulk density ( $\text{g cm}^{-3}$ ).

Four replicates were completed for each pest, formulation and concentration. Statistical analyses were performed using SAS Stat Software v8.02. The Gauss–Newton method was used for non-linear regressions to estimate the  $C_0$ ,  $C_{50}$ ,  $a$  and  $b$  parameters of the survival model (eqn (1)).  $\text{LD}_{50}$  and  $\text{LD}_{90}$  values were estimated with a bootstrap method using 1000 simulations and residuals as a source of variation.<sup>23</sup>

This explains the similar but slightly different values for  $C_{50}$  and  $\text{LD}_{50}$ . If there were only one curve or no variation between replicates and formulations, the values of  $C_{50}$  and  $\text{LD}_{50}$  would have been exactly the same. Comparisons between formulations were completed based on the bootstrap approach results<sup>23</sup> and indicated that only the nematodes were sensitive to formulation.

## 2.3 Soil management

The materials and methods were similar to Allaire *et al.*<sup>15</sup> In short, an  $0.6 \times 0.6 \times 0.025 \text{ m}$  acrylic column was packed by hand to a density of  $1.6 \text{ g cm}^{-3}$  with the same soil as used for the previous experiment. This high soil density is regularly observed in B horizons. The soil surface reproduced a bed-furrow system centered in the middle of the column (Fig 1). The bed was  $0.12 \text{ m}$  high and the bed top was  $0.3 \text{ m}$ . A high-density polyethylene plastic film (Triscal, Hollister, CA) partially covered the surface. It covered only the bed, not the furrow. It is common practice in Southern California to install such a tarp cover for strawberry, pepper and tomato production right after fumigation. The soil column was gastight at its lower and side boundaries. One milliliter of liquid 3BP (97% pure; Fluka) was injected in the center of the column (about  $1000 \text{ kg ha}^{-1}$ )  $0.3 \text{ m}$  below the bed surface and  $0.3 \text{ m}$  from the left side of the column. This application rate is much higher than we would expect for field application since it corresponds roughly to 6, 16 and 42 times the maximal applied dose (dose much higher than needed to control all three pests) used in the previous experiment for fungus, grass and nematode, respectively. This high application rate was chosen to increase the accuracy of concentration measurements in the profile, to accelerate the trial and because high and low dose would result in the same distribution pattern trends. Volatilization chambers were installed



**Figure 1.** Schematic representation of the bed-furrow system and distribution of the sampling points.

at the soil surface to circulate fresh air into the chamber. The volatilization flux was measured with a system of valves connected to a gas chromatograph.

The experiment was set up to explore the effect of combining the partial covering of the soil and different furrow irrigation managements on an initially moist soil ( $0.17 \text{ cm}^3 \text{ cm}^{-3}$  of water or 60% field capacity), similar to the previous experiment. Irrigation treatments were as follows: (1) No irrigation (M-0). (2) A 5-h furrow irrigation (a total of 0.9 liter on each side, roughly equivalent to an 8-cm irrigation depth distributed over an entire field) was applied 24 h after injection (M-5). This was applied by maintaining a 3-cm pressure head at the soil surface using a Mariotte bottle setup. The duration was long due to the high soil density. (3) A 2-h furrow irrigation was applied 2 h after injection and repeated every 24 h thereafter for four consecutive days (M-M). A total of 0.6 liter water was added to each side using a Mariotte bottle set-up. This represents roughly a series of 1.3 cm depth irrigations distributed over an entire field for sealing the soil surface.

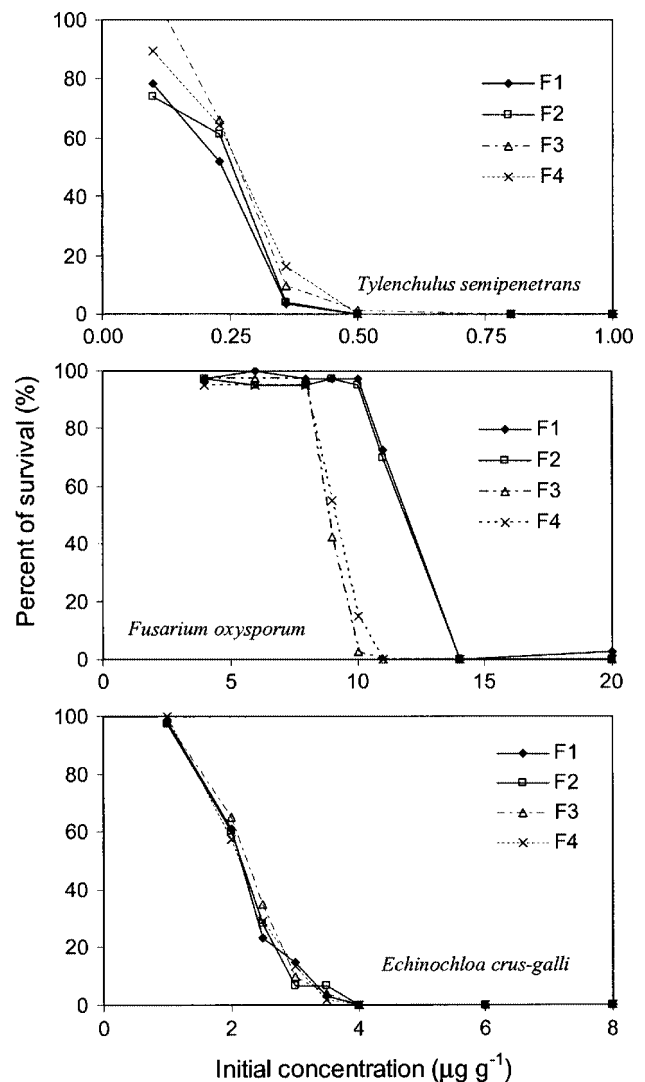
3BP concentration in the gas phase ( $C_g$ ) was measured at 62 different sites (Fig 1) in the column several times during the first 2 days, then once a day for up to 9 days. Fifty microliters of soil air was sampled and transferred into headspace vials following the methods of Allaire *et al.*<sup>15</sup> 3BP concentration was determined using gas chromatography following the method of Gan *et al.*<sup>24</sup>

Temporal interpolation of concentration values was completed with a least-square regression method so that CT could be estimated at each sampling site. Variograms were developed at each time of interest for spatial interpolation of CT values. Ratio and angle of anisotropy, model, nugget, tolerance and direction of the variograms were used for kriging interpolation using Surfer8 software (Golden Software Inc). Variograms indicate good correlation between concentrations at different distances and anisotropy was negligible. Spatial correlation between CT values was well defined. Thus, interpolation was quite accurate. The percentage of the profile that had reached  $CT_{90}$  was then estimated at different times for each pest using the same software.

### 3 RESULTS AND DISCUSSION

#### 3.1 Model

The shape of the response curve to 3BP was similar for the three species (Fig 2) and regression always converged with good accuracy (1% error). The model was therefore accurate for describing the pest response to fumigation. Statistical analysis indicated that  $C_0$  was not significantly different from zero for all but two regressions (two formulations on the fungi). A minimal threshold value of zero ( $C_0$ ) was also obtained for methyl bromide and for different pests.<sup>25</sup> A large  $b$  value indicates a steeper slope, ie a small change in concentration has a large effect on pest control. Nematode



**Figure 2.** *Fusarium oxysporum*, *Echinochloa crus-galli* and *Tylenchulus semipenetrans* (fungus, grass and nematode) survival as a function of total 3BP fumigation rate and formulation. The points are average measured values of four replicates.

and grass have similar  $b$  values while the fungus has a much higher  $b$  value (Table 2). Therefore, although more fumigant was required to control the impact of fungus (Table 2), a slight change of 1% in concentration was enough to change from  $LD_{50}$  to  $LD_{90}$ , while an increase of 150% was required for grass. Thus, the dose for fungus seems trickier to adjust.

#### 3.2 Lethal dosage

Comparing literature results is difficult because different methodologies have been used to obtain and analyze information. Often, concentrations cannot be transformed from one phase to another because various soil parameters such as density or water content are missing. For this reason, the indexes are presented on the basis of different phases.

The lethal dose for controlling 50% of the pest population ( $LD_{50}$ ), based on the total applied mass, was  $10 \mu\text{g g}^{-1}$  soil weight for fungus, which is nearly three times higher than for grass and 30 times higher than for nematode (Table 3).  $LD_{50}$  values for the

**Table 2.** Non-linear regression parameters ( $a$ ,  $C_{50}$  and  $b$ , eqn (2), Section 2.2)<sup>a</sup> describing survival of *Tylenchulus semipenetrans*, *Fusarium oxysporum* and *Echinochloa crus-galli* (nematode, fungus and grass) in response to propargyl bromide fumigation dose based on total, gas or liquid phases

Pest	Total			Gas			Liquid		
	$a^a$ ( $\mu\text{g g}^{-1}$ )	$C_{50}$ ( $\mu\text{g g}^{-1}$ )	$b$ —	$a$ ( $\mu\text{g cm}^{-3}$ )	$C_{50}$ ( $\mu\text{g cm}^{-3}$ )	$b$ —	$a$ ( $\mu\text{g cm}^{-3}$ )	$C_{50}$ ( $\mu\text{g cm}^{-3}$ )	$b$ —
Nematode	80.8	0.251	8.46	78.6	0.022	8.92	78.92	0.463	5.80
Fungus	98.1	11.59	22.3	98.8	1.02	28.1	97.02	20.5	22.7
Grass	95.8	2.20	8.37	96.3	0.191	7.56	96.7	3.95	7.15

<sup>a</sup> Parameters  $a$ ,  $C_{50}$ , and  $b$  were always significant at  $P < 0.0001$  while  $C_0$  was not significant at  $P > 0.05$ . The latter is not presented in this table. Only the results of the most efficient formulation are presented.

**Table 3.** Lethal dose ( $LD_{50}$ ,  $LD_{90}$ ) and concentration-time index ( $CT_{90}$ ) for controlling *Tylenchulus semipenetrans*, *Fusarium oxysporum* and *Echinochloa crus-galli* (nematode, fungus, and grass) based on total, gas and liquid concentrations when fumigated with propargyl bromide

Pest	Total			Gas			Liquid		
	$LD_{50}$ ( $\mu\text{g g}^{-1}$ )	$LD_{90}$ ( $\mu\text{g g}^{-1}$ )	$CT_{90}^a$ ( $\mu\text{g h g}^{-1}$ )	$LD_{50}$ ( $\mu\text{g cm}^{-3}$ )	$LD_{90}$ ( $\mu\text{g cm}^{-3}$ )	$CT_{90}$ ( $\mu\text{g h cm}^{-3}$ )	$LD_{50}$ ( $\mu\text{g cm}^{-3}$ )	$LD_{90}$ ( $\mu\text{g cm}^{-3}$ )	$CT_{90}$ ( $\mu\text{g h cm}^{-3}$ )
Nematode	0.232 ( $\pm 0.004$ )	0.304 ( $\pm 0.024$ )	11.2	0.021 ( $\pm 0.0004$ )	0.027 ( $\pm 0.002$ )	1.00	0.419 ( $\pm 0.008$ )	0.565 ( $\pm 0.020$ )	20.8
Fungus	11.3 ( $\pm 0.2$ )	12.0 ( $\pm 0.6$ )	439	1.01 ( $\pm 0.01$ )	1.07 ( $\pm 0.06$ )	39.4	20.2 ( $\pm 0.3$ )	21.3 ( $\pm 1.1$ )	782
Grass	2.14 ( $\pm 0.05$ )	2.98 ( $\pm 0.10$ )	109	0.193 ( $\pm 0.003$ )	0.266 ( $\pm 0.009$ )	9.77	3.86 ( $\pm 0.08$ )	5.34 ( $\pm 0.21$ )	196

<sup>a</sup> The concentration-time ( $CT_{90}$ ) index was directly calculated from  $LD_{90}$ . Therefore, the error is directly associated with  $LD_{90}$ .

same grass species correspond to those of Ma *et al*<sup>2</sup> who found a value of  $2.8 \mu\text{g g}^{-1}$  for the same soil. These values correspond to a  $CT_{90}$  of  $345 \mu\text{g g}^{-1} \text{ h}$  for fungus (Table 3),  $109 \mu\text{g g}^{-1} \text{ h}$  for grass and  $11 \mu\text{g g}^{-1} \text{ h}$  for nematode. Thus, if all three species are present in the same soil, the dosage for controlling the impact of the fungus on plant growth should fully control the two other pests.

Based on the liquid phase,  $LD_{50}$  and  $LD_{90}$  values were slightly higher (5–10%) than those of Ma *et al*<sup>2</sup> for grass and fungus. The differences in the values may be due to experimental differences and, possibly, the fact that they used 97% pure 3BP. Indeed, it appears that formulation influences the efficiency of fumigation. The nematode was the most sensitive to the formulation. The  $LD_{90}$  value, based on the total applied

mass, ranged from  $0.3 \mu\text{g g}^{-1}$  when treated with the most efficient formulation, to  $0.4 \mu\text{g g}^{-1}$  when treated with the least efficient formulations. The fungus was found to be slightly less sensitive to 3BP formulation, followed by grass for which the  $LD_{90}$  values were not significantly different between formulations (Fig 2).

Results found in the literature from field studies and laboratory experiments showed 3BP dose values in the same range as those used for this study. A good control of *F. oxysporum* was obtained with  $90 \text{ kg ha}^{-1}$  in a field experiment with a tarped soil and  $98 \text{ kg ha}^{-1}$  in a closed system (Table 4), while  $LD_{90}$  in this experiment corresponds to  $78\text{--}94 \text{ kg ha}^{-1}$  depending on the formulation.

The values for controlling nematodes (*T. semipenetrans* or *Meloidogyne* sp) in different field studies range

**Table 4.** Results from different studies on controlling pests using alternative fumigants

Fumigant	Pests	Dose ( $\text{kg ha}^{-1}$ ) <sup>a</sup>	Soil and conditions	Reference
3BP	<i>Tylenchulus semipenetrans</i> and <i>Meloidogyne</i> sp	200	Field, drip applied	33
3BP	<i>Tylenchulus semipenetrans</i> and <i>Meloidogyne</i> sp	224	Field, shank applied, tarped	33
3BP	<i>Meloidogyne</i> sp	45	Field, tarped	34
3BP	<i>Meloidogyne</i> sp	168	Field	35
3BP	<i>Fusarium oxysporum</i>	98	Sandy loam, closed system	2
3BP	<i>Fusarium oxysporum</i>	90	Field, tarped	35
3BP	<i>Echinochloa crus-galli</i>	32	Sandy loam, closed system	2
Methyl iodide	<i>Heterodera schachtii</i>	38	Sand, laboratory	14
1,3-Dichloropropene	<i>Tylenchulus semipenetrans</i>	112	Sandy loam, small field plots	26
Metam-sodium	<i>Fusarium</i> spp	1400	Field, tarped	27
1,3-Dichloropropene + chloropicrin (60:40)	?	278	Field, tarped	36

<sup>a</sup> Best dose when higher application rates did not result in better pest control or gave yields similar or higher than methyl bromide.

between 45 and 224 kg ha<sup>-1</sup> (Table 4). This is much higher than the values found in this study (3.1 kg ha<sup>-1</sup>). The effective dose seems to vary depending on soil, method of application, soil tarping<sup>3</sup> (Table 4) and formulation. In addition to these factors, the difference from our study may be due to the higher efficiency of closed systems compared with field studies<sup>26</sup> and to the fact that efficiency in this study was based on active motile nematodes. Cyst nematodes are often found in southern California during dry conditions. Cysts tend to be more resistant than active nematodes to fumigants.<sup>26</sup> Considering that fumigation in Southern California is often applied during dry conditions, LD<sub>90</sub> may not accurately estimate the actual control when cysts are present. In our work, non-motile nematodes were considered dead. Some of them might have been still alive and infectious.<sup>12</sup> Therefore, the dose required to kill the nematode population could be underestimated in this study.

CT<sub>90</sub> for fungus based on the gas phase was 1.0 µg cm<sup>-3</sup> h in this study compared with 12 µg cm<sup>-3</sup> h when fumigated with 1,3-dichloropropene for the same species and the same soil.<sup>10</sup> A dose of 112 kg ha<sup>-1</sup> was required to kill *T semipenetrans* with 1,3-dichloropropene in small field plots.<sup>27</sup> Using a closed system, Becker *et al*<sup>26</sup> found LD<sub>90</sub> values for *T semipenetrans* ranging from 21 to 837 kg ha<sup>-1</sup> when fumigated with methyl bromide and methyl iodide. In another study,<sup>9</sup> *F oxysporum* was controlled in field trials using 300 kg ha<sup>-1</sup> of methyl bromide. Other comparisons are given in Table 4. These values tend

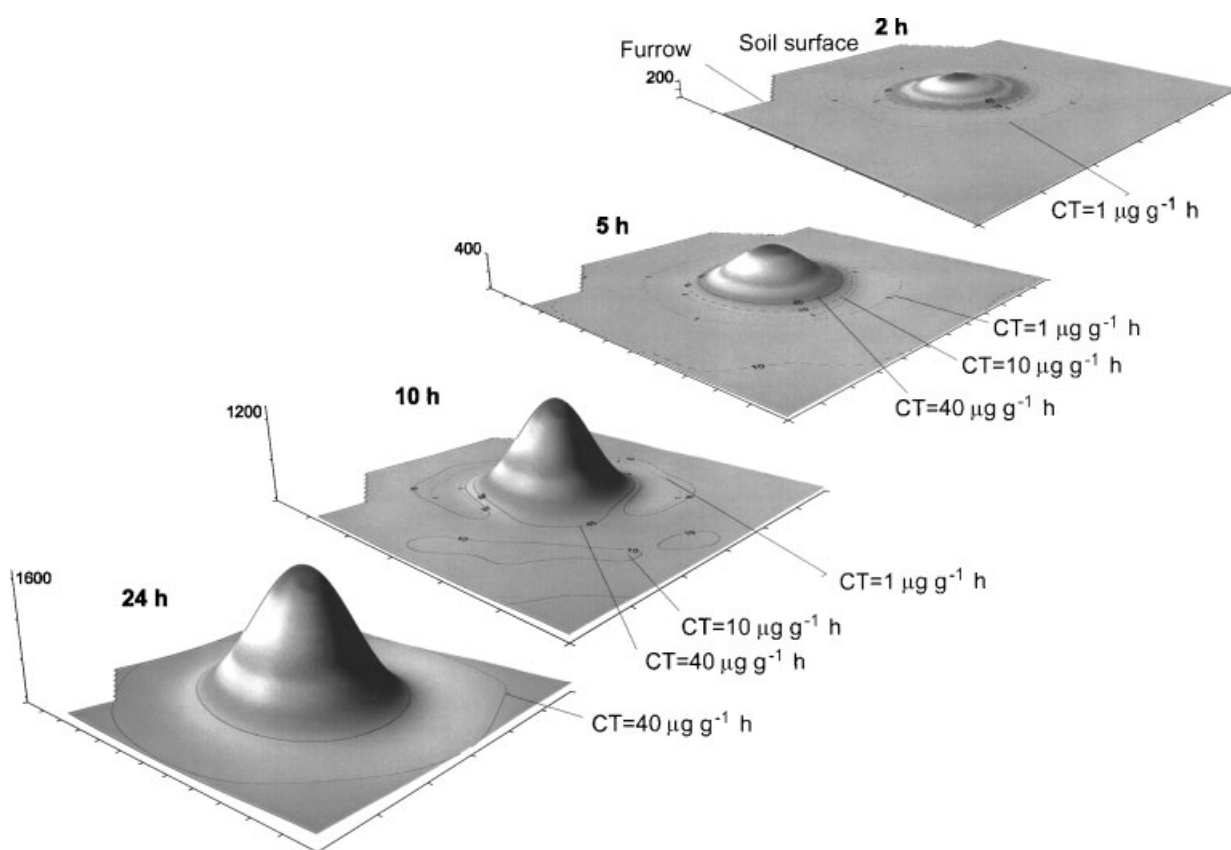
to indicate that 3BP is as efficient in controlling different pests as methyl bromide, methyl iodide or 1,3-dichloropropene.

### 3.3 Soil management

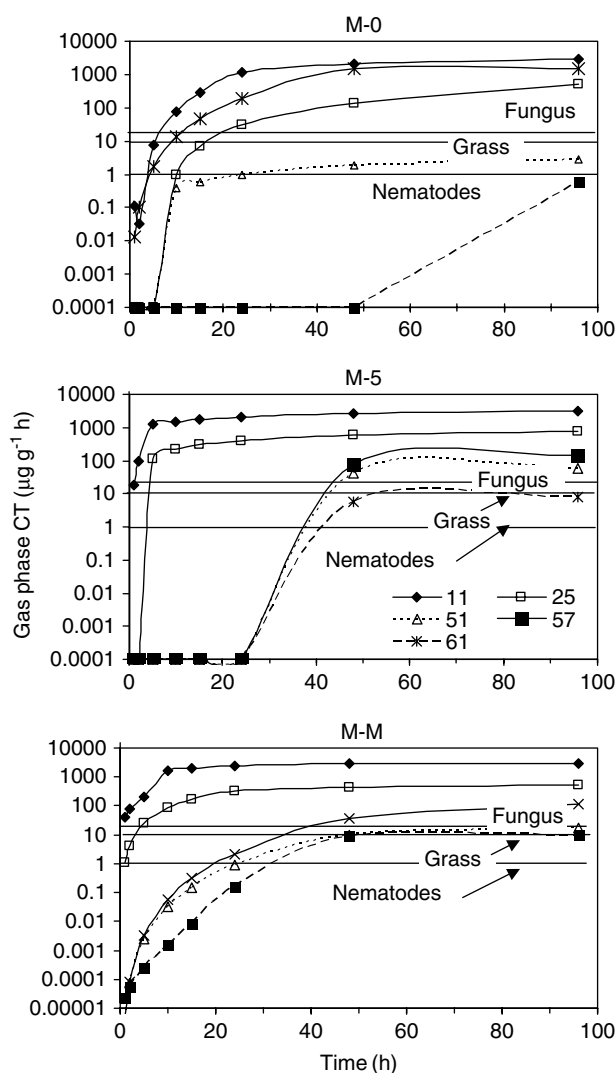
#### 3.3.1 CT distribution in time and space

Distribution of the gas-phase CT values in the profile is plotted for different times in Fig 3. Figure 4 shows the gas-phase CT as a function of time at several points in the profile for three treatments (for the exact location of points, refer to Fig 1).

CT distribution expanded radially from the injection point at all times before irrigation (Fig 3). CT values varied over nine orders of magnitude within 96 h. Close to the injection (sampling point 11), CT<sub>90</sub> was reached for all three species within a few hours in the irrigated treatments (Fig 4, M-5 and M-M). Further from the injection point (sampling point 25), it took 2 to 3 times longer to control 90% of the pest population (CT<sub>90</sub>) depending upon the treatment. Even further from the injection point (point 57 at the lower boundary), it took between 16–36 h to control nematode, 36–38 h to control grass and 39–40 h to control fungus in the irrigated treatments (M-5 and M-M). Comparatively, the bottom boundary of non-irrigated treatment (M-0) did not reach CT<sub>90</sub> for any species during the same time period (Fig 4). Near the surface below the tarped bed (sampling point 51), the nematodes were controlled within 10 h in the M-0 treatment. Grass and fungus never reached CT<sub>90</sub> in M-0 treatment. Comparatively, it took roughly 40 h to



**Figure 3.** Gas-phase CT distribution in the soil profile at different times in the treatment with a 2-h furrow irrigation repeated four times (M-M)..



**Figure 4.** Gas phase CT values at different points in the profile under different irrigation treatments. Horizontal lines indicate CT<sub>90</sub> index for *Fusarium oxysporum*, *Echinochloa crus-galli*, and *Tylenchulus semipenetrans*, respectively (fungus, grass and nematode).

control all species in M-5 and M-M (Fig 4). This is important considering the high application rate compared to what would be expected in the field. It implies that, with lower application rate (about 10-fold), it would take at least ten times longer to reach CT<sub>90</sub> at the injection point and even longer away from it because the fumigant would have more time to degrade and volatilize.

Therefore, the control zone is expected to be significantly smaller in the field than observed in this experiment. Even if a good control were obtained in the root zone of a homogeneous soil, a certain number of pests will remain alive at some locations, such as near the soil surface and at deeper depths, and would be a potential source for recolonisation. Considering that 25–50% of young strawberry roots are in the top 7.5 cm of soil,<sup>28</sup> there is a high risk of recolonisation of the roots from the surface even if the bed is covered with a tarp and the furrow is sealed with water. This problem significantly increases in the field where heterogeneity of soil properties, water

**Table 5.** Percentage of the soil profile<sup>a</sup> that has reached the gas phase concentration–time index (CT<sub>90</sub>) under different treatments at different times for *Tylenchulus semipenetrans*, *Fusarium oxysporum* and *Echinochloa crus-galli* (nematode, fungus and grass)

Treatment	Time (h)	Nematode	Fungus	Grass
M-0	2	82	6	9
	5	82	6	36
	10	85	12	76
	24	85	70	79
	96	>92	73	79
M-5	2	82	9	48
	5	88	9	88
	10	91	76	91
	24	91	82	91
	96	>92	>92	>92
M-M	2	61	6	9
	5	88	12	24
	10	88	18	79
	24	91	48	79
	96	>92	>92	>92

<sup>a</sup> The total area of the soil profile underneath soil surface was 0.33 m<sup>2</sup>.

distribution and pest distribution is much higher and where surface sealing with tarp and/or water is not used.

California code of regulation (Title 3, Division 6, Chapter 2, Subchapter 4, Article 4)<sup>29</sup> requires that a soil undergoing shallow injection in a bed-furrow system must keep the tarp on the bed for at least 5 days following methyl bromide injection. This regulation was primarily written to improve environmental protection and safety, but it also helps to increase the efficiency of fumigation. From the point of view of efficiency, the tarp should be kept on the soil surface for 3BP at least as long as for methyl bromide if fungus has to be controlled. This is explained by the fact that it took at least 96 h to control this pest in more than 90% of the soil profile (Table 5) under a very high application rate, and the fact that irrigation, convection, soil degradation, total losses, and dissipation are more variable and more important in the field.

### 3.3.2 Effect of irrigation

It is often considered that fumigation of relatively dry soil leads to better pest control because more of the pore space is available for gas movement<sup>3</sup> and diffusion in air of most gases is about 10 000 times faster than diffusion in water.<sup>30</sup> This commonly held belief is not fully supported by the results shown herein. Based on measured gas-phase concentrations, more than 92% of the profile was controlled for fungus and grass in M-M treatment while less than 80% was controlled in M-0 treatment 96 h after fumigation (Table 5). This results from the fact that losses, mainly due to volatilization, in M-0 (21%) were much higher than in M-M treatment (6%) resulting in a



lower mass of chemical in the profile. The highest degradation, mainly due to hydrolysis, that occurred in M-M (51%) compared with M-0 (32%), was not sufficient to counterbalance the volatilisation effect on 3BP residual mass in the profile<sup>18</sup>. M-5 treatment seemed more efficient right after fumigation than the two other treatments, but tended toward the M-M treatment at later time in terms of pest control (Table 5).

Therefore, an approach using multiple irrigations has the advantage of decreasing volatilization, requiring 1/3 less water<sup>18</sup> and offers better pest control because a higher portion of the profile reaches CT<sub>90</sub>. However, it is important to keep in mind that the effective phase of 3BP for the different pests is unknown.

#### 4 CONCLUSION

LD<sub>50</sub> based on the total applied mass was about 0.23, 2.1 and 11 µg g<sup>-1</sup> for a nematode, a grass and a fungus, respectively in a sandy loam soil. LD<sub>90</sub> values were 0.3, 3.0 and 12 µg g<sup>-1</sup> and CT<sub>90</sub> values 11, 109 and 345 µg g<sup>-1</sup> h for *Tylenchulus semipenetrans*, *Echinochloa crus-galli* and *Fusarium oxysporum*, respectively. 3BP seems as efficient as other alternative fumigants in controlling these three species. Irrigation influenced the time required to control pests because it had an impact on gas distribution, volatilization and degradation in the profile. CT<sub>90</sub> for the most resistant pest was not reached at the lower end of the column and near the surface in the non-irrigated treatment even though the bed was covered with tarp and a high fumigation rate was applied. Multiple short irrigations tended to show better pest control because they control a larger soil volume. More research is needed to develop recommendations regarding field application for other species, soil types, and management techniques.

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